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EXAMINER	
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Please find below and/or attached an Office communication concerning this application or proceeding.

The time period for reply, if any, is set in the attached communication.

Office Action Summary	Application No.	Applicant(s)	
	10/531,626	GAMBLE ET AL.	
	Examiner	Art Unit	
	Magdalene K. Sgagias	1632	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE *3* MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *01 October 2009*.

2a)☒ This action is **FINAL**. 2b)☐ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-6,8,10,13,15,21-26 and 43* is/are pending in the application.

4a) Of the above claim(s) *21-24 and 26* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *1-6,8,10,13,15,25 and 43* is/are rejected.

7)☐ Claim(s) _____ is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☐ The specification is objected to by the Examiner.

10)☒ The drawing(s) filed on *14 April 2005* is/are: a)☒ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☐ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☐ All b)☐ Some * c)☐ None of:

1.☐ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☐ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☐ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date _____
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____.
5) ☐ Notice of Informal Patent Application
6) ☐ Other: _____.

DETAILED ACTION

Applicant's arguments filed 10/01/2009 have been fully considered. The amendment has been entered. Claims 1-6, 8, 10, 13, 15, 21-26, 43 are pending. Claims 7, 9, 11-12, 14, 16-20, 27-42 are canceled. Claims 21-24, 26 are withdrawn. Claims 1-6, 8, 10, 13, 15, 25, 43 are under consideration.

Claim Rejections - 35 USC § 112, 1st paragraph

The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claims 1-6, 8, 10, 13, 15, 25, 43 **remain** rejected under 35 U.S.C. 112, first paragraph, as containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

The claims embrace an enormous number of sphingosine kinase functional fragments or homologs, constituting a claimed genus. The specification fails to disclose a representative number of the numerous sphingosine kinase nucleic acid encoding fragments or homologs thereof, of any size sequence that would be able to modulate one or more mammalian endothelial cell functional characteristics in vivo. The specification does not describe the structure or functional nature of the numerous nucleic acid fragments or homologs thereof encoding sphingosine kinase that will modulate one or more mammalian endothelial cell functional characteristics in vivo. The specification is further silent on the specific characteristics, or sequence motifs of sphingosine kinase or homolog thereof that may contribute to a

therapeutic treatment and/or prophylaxis characterized by aberrant or otherwise unwanted endothelial cell functioning in a mammal. The claims thus embrace a claimed genus that encompasses nucleic acid sequences or homologs thereof, yet to be discovered. As the specification fails to disclose any sphingosine kinase nucleic acid sequences or homologs thereof, the Artisan of skill could not predict that Applicant possessed any species of said sphingosine kinase nucleic acid sequences. To satisfy the written description requirement, a patent specification must describe the claimed invention in sufficient detail such that the Artisan can reasonably conclude that the inventor(s) had possession of the claimed invention. Such possession may be demonstrated by describing the claimed invention with all of its limitations using such descriptive means as words, structures, figures, diagrams, and/or formulae that fully set forth the claimed invention. Possession may be shown by an actual reduction to practice, showing that the invention was "ready for patenting", or by describing distinguishing identifying characteristics sufficient to show that Applicant was in possession of the claimed invention (January 5, 2001 Fed. Reg., Vol. 66, No. 4, pp. 1099-11). Moreover, MPEP 2163 states:

> [A] biomolecule sequence described only by a functional characteristic, without any known or disclosed correlation between that function and the structure of the sequence, normally is not a sufficient identifying characteristic for written description purposes, even when accompanied by a method of obtaining the claimed sequence.
>
> Applicant's attention is also directed to *In re Shokal*, 113 USPQ 283 (CCPA 1957), wherein it is stated:
> It appears to be well settled that a single species can rarely, if ever, afford sufficient support for a generic claim. *In re Soll*, 25 CCPA (Patents) 1309, 97 F2d 623, 38 USPQ 189; *In re Wahlforss*, 28 CCPA (Patents) 867, 117 F2d 270, 48 USPQ 397. The decisions do not however fix any definite number of species which will establish completion of a generic invention and it seems evident therefrom that such number will vary, depending on the circumstances of particular cases. Thus, in the case of small genus such as the halogens, consisting of four species, a reduction to practice of three, perhaps even two, might serve to complete the generic invention, while in the case of a genus comprising hundreds of species, a considerably larger number of reductions to practice would probably be necessary.

Overall, what these statements indicate is that the Applicant must provide adequate description of such core structure and function related to that core structure such that the Artisan of skill could determine the desired effect. Hence, the analysis above demonstrates that Applicant has not determined the core structure for full scope of the claimed genus. In analyzing whether the written description requirement is met for genus claims, it is first determined whether a representative number of species have been described by their complete structure. Therefore, the breadth of the claims as reading on numerous sphingosine kinase nucleic acid functional fragment sequences or homologs thereof, yet to be discovered; in view of the level of knowledge or skill in the art at the time of the invention, an Artisan of skill would not recognize from the disclosure that Applicant was in possession of the genus of nucleic acid sequences encoding sphingosine kinase motifs with mammalian endothelial cell functional characteristics in vivo. Thus it is concluded that the written description requirement is not satisfied for the claimed genus. In conclusion, this limited information is not deemed sufficient to reasonably convey to one skilled in the art that Applicant is in possession of numerous therapeutic nucleic acid fragments of homologs thereof sequence motif, at the time the application was filed. Thus it is concluded that the written description requirement is not satisfied for the claimed genus.

A. Applicants argue what is conventional or well known to one of ordinary skill in the art need not be disclosed in detail. Applicants respectfully submit that sphingosine kinase sequences, fragments, and homologs thereof were amply described in the art at the time the instant application was filed. For example, U.S. Patent No. 6,730,480 ("the "480 patent"), with a priority date of May 13, 1999, discloses human sphingosine kinase-1 polypeptides, polynucleotides, and assays for measuring sphingosine kinase-1 activity. The first issued claim in "480 patent recites: 1. An isolated polypeptide comprising an amino acid sequence at least

90% identical to the sequence of SEQ ID NO: 2 [human sphingosine kinase-1] wherein said polypeptide has sphingosine kinase activity. Thus, Applicants were in possession of all sphingosine kinases disclosed and claimed in the "480 patent at the time the instant application was filed. In addition, U.S. Patent No. 7,112,427 ("the "427 patent"), with a priority date of May 13, 1999, discloses human sphingosine kinase-1 polypeptides, polynucleotides, and assays for measuring sphingosine kinase-1 activity. The first issued claim in "427 patent recites: 1. An isolated polynucleotide encoding a sphingosine kinase, the polynucleotide comprising (1) the sequence of SEQ ID NO:I [human sphingosine kinase-1], (2) a sequence at least 90% identical to SEQ ID NO: 1, (3) a nucleotide sequence that hybridizes to the nucleotide sequence of (1) or (2) under high stringency conditions of about 65.degree. C. and about 50% v/v formamide and about 0.15M salt, (4) a nucleotide sequence encoding a polypeptide having the sequence of SEQ ID NO:2 [human sphingosine kinase-1], or (5) a nucleotide sequence complementary to the nucleotide sequence of any one of(l) to (4). Thus, Applicants were in possession of all sphingosine kinases disclosed and claimed in the "427 patent at the time the instant application was filed. Furthermore, Pitson et al., Biochem J. Sep 1;350 Pt 2:pp. 429-41, 2000a, (of record) disclose homologs of the human sphingosine-1 kinase in mouse, rat, monkey, S. cerevisiae, S. pombe, A. thaliana, and O. sativa and evolutionarily conserved sequences thereof. Kohama et al., 1998 (of record) disclose two murine sphingosine kinase one splice variants, evolutionary conserved sequences compared to human, yeast, and nematode sphingosine-1 kinases, and sphingosine kinase activity assays (see, for example Figures 5 and 6 of Kohama). Pitson et al., 2002 (of record) disclose no less than 14 human sphingosine kinase-1 mutants that have reduced sphingosine kinase activity (see, for example Figure 3 and Table 1). Pitson et al. (The Journal of Biological Chemistry. Vol. 275, No. 43, pp. 33945-33950, 2000b; copy attached) disclose site-directed mutagenesis of Gly82 to Asp (G82D) of the human sphingosine kinase-1.

The residue was identified through sequence conservation between several diacylglycerol kinases and sphingosine kinases (see, for example, Figure 1). The G82D mutation acts as a dominant negative to block activation of sphingosine kinase-1 activity and sphingosine kinase-1 cell signaling. In addition, the abstract of Pitson et al. (FEBS Lett. 2001 Dec 7; 509(2):169-73; abstract attached) discloses a human sphingosine kinase 1 point mutant, Gly113 to Ala (G113A), with increased catalytic activity. The residue was identified through sequence conservation.

These arguments are not persuasive because all of the above cited references fail to indicate that applicants were in possession of any representative number of the numerous sphingosine kinase nucleic acid encoding fragments or homologs thereof, of any size sequence that would be able to modulate one or more mammalian endothelial cell functional characteristics in vivo. The issue is not the possession of the sphingosine kinase nucleic acid sequences per se as cited in the above references which exhibit sphingosine kinase activity in vitro. The issue is the possession of the numerous sphingosine kinase nucleic acid encoding fragments or homologs thereof, of any size sequence that would be able to modulate one or more mammalian endothelial cell functional characteristics in vivo. For example, the "480 patent, discloses human sphingosine kinase-1 polypeptides, polynucleotides, and assays for measuring sphingosine kinase-1 activity, however, fails to provide possession of the numerous sphingosine kinase nucleic acid sequences encoding sphingosine kinase in vivo that would result in modulating on or more mammalian endothelial cell functional characteristics in vivo, as instantly claimed.

B. Applicants argue that the claims are directed to methods of modulating one or more mammalian endothelial cell functional characteristics, the method comprising inducing over-expression of sphingosine kinase, functional fragment thereof, or homolog thereof having

sphingosine kinase activity. One having skill in the art would appreciate that the claims clearly recite sphingosine kinases having sphingosine kinase activity which is an important aspect of the therapeutic treatment and/or prophylaxis (see, for example, Figures 11, 15, and 16 and Example 2). Further, the Action alleges that the specification does not provide any teachings whether such fragment would retain the function of sphingosine kinase and that there is no identification of any particular portion of the structure that must be conserved. Applicants respectfully disagree. As noted above, Pitson et al., 2001 and Pitson et al., 2001 provide examples of highly conserved regions in sphingosine kinase type I kinases that were mutated to either inhibit or increase sphingosine kinase activity. One having skill in the art would appreciate that this is a standard method in the art, as sequences conserved across evolution are thought to be functionally important sequences. Thus, Applicants submit that it is the highly conserved regions that inform where functional portions of the molecule are located and where mutations could be made. Accordingly, the U.S.P.T.O. believes Applicants to be in possession of a considerably large genus of sphingosine kinase-1 molecules (see, U.S. Patent Nos. 6,730,480 and 7,112,427) and the art has provided examples of homologs, functional fragments, and methods of identifying the same (see, for example Kohama et al., 1998, Pitson et al., 2000a, Pitson et al., 2000b, and Pitson et al., 2001). Thus, one having skill in the art would reasonably conclude that Applicants were in possession of the presently claimed genus of sphingosine kinases at the time the application was filed.

These arguments are not persuasive because recognizing the importance of sphingosine kinase activity for therapeutic treatment and/or prophylaxis fails to provide possession of the numerous sphingosine kinase nucleic acid sequences encoding sphingosine kinase resulting in modulating one or more mammalian endothelial cell characteristics in vivo. For example, Pitson et al as cited above provide examples of highly conserved regions in

sphingosine kinase type I kinases that were mutated to either inhibit or increase sphingosine kinase activity, however these conserved regions fail to provide possession by the applicant of sphingosine kinase nucleic acid sequences exhibiting any endothelial cell functional characteristics in vivo. Thus, Applicants submit that it is the highly conserved regions that inform where functional portions of the molecule are located and where mutations could be made. While the specification discloses that the sphingosine kinase homologues all possess an extremely high degree of sequence identity in the nucleotide binding domain and knowing how to generate fragments parts or fragments however, an artisan would not know what to mutate and where to mutate the gene. The claimed invention is not adequately described if the claims require essential or critical functional fragment or homologues of sphingosine kinase nucleic acid sequences which would result in modulating one or more mammalian endothelial cell functional characeristics. Possession may be shown by actual reduction to practice, or by describing the invention with sufficient relevant identifying characteristics as it relates to the claimed invention as a whole such that one of skill in the art would recognize that Applicants had possession of the invention. The specification does not provide any teachings whether such fragment would retain the function of sphingosine kinase. To provide evidence of possession of a claimed genus, the specification must provide sufficient distinguishing identifying characteristics of the genus. The factors to be considered include disclosure of complete or partial structure, physical and/or chemical properties, functional characteristics, and structure/function correlation, methods of making the claimed product or any combination thereof. In this case, there is no identification of any particular portion of the structure that must be conserved. Accordingly, in the absence of sufficient recitation of distinguishing identifying characteristics, the specification does not provide written description of the claimed genus. The claims are extremely broad since insufficient guidance is provided as to which of the plethora of

fragments of nucleic acids encode sphingosine kinase polypeptides which will retain the characteristics of a functional sphingosine kinase. While the claims are directed to fragment of nucleic acid encoding sphingosine kinase Applicants do not disclose any actual examples or prophetic examples on expected performance parameters of any of possible encoded fragments of a functional sphingosine kinase. Second a skilled artisan having knowledge of the mouse, rat, monkey, S. cerevisiae, S. pombe, A. thaliana, and O. saliva sphingosine kinase sequences and that the sphingosine kinase homologues all possess an extremely high degree of sequence identity in the nucleotide binding domain and knowing how to generate fragments of the sphingosine kinase molecule will not be able to recognize that the Applicant was in possession of the claimed invention because each fragment having distinct sequence structure elicit distinct functional characteristics in the claimed sphingosine kinase molecule. The broadly claimed fragments fail to uniquely identify the structural and functional characteristics of the claimed sphingosine kinase.

Claims **1-6, 8, 10, 13, 15, 25, 43 remain** rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for overexpression of sphingosine kinase by introducing into mammalian endothelial cells a nucleic acid encoding sphingosine kinase resulting in enhancing cell survival, altering adhesion molecule expression and enhancing neutrophil adhesion to endothelial cells and promote tube formation or the endothelial cells arrange into capillary like network in vitro, does not reasonably provide enablement for modulating one or more mammalian endothelial cell functional characteristics by way of the claimed methods in vivo. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the invention commensurate in scope with these claims.

Independent claim 1 embraces a method of modulating one or more mammalian endothelial cell functional characteristics, said method comprising inducing over-expression of sphingosine kinase, wherein said over-expression is achieved by introducing into said endothelial cell an isolated nucleic acid molecule encoding sphingosine kinase or functional fragment or homolog thereof, wherein said kinase functional fragment thereof, or homolog, thereof comprises sphingosine kinase activity. Independent claim 2 embraces a method of modulating one or more endothelial cell functional characteristics in a mammal, said method comprising inducing over-expression of sphingosine kinase, wherein said over-expression is achieved by introducing into said endothelial cell am isolated nucleic acid molecule encoding sphingosine kinase or functional fragment or homolog thereof, wherein said kinase functional fragment thereof, or homolog, thereof comprises sphingosine kinase activity. Independent claim 3 embraces a method for the treatment and/or prophylaxis of a condition characterized by aberrant or otherwise unwanted endothelial cell functioning in a mammal, said method comprising inducing over-expression of sphingosine kinase, wherein said over-expression is achieved by introducing into said endothelial cell an isolated nucleic acid molecule encoding sphingosine kinase or functional fragment or homolog thereof, wherein said kinase functional fragment thereof, or homolog, thereof comprises sphingosine kinase activity.

The specification teaches that overexpression of sphingosine kinase by introducing an adenovirus containing sphingosine kinase enhances cell survival of human umbilical vein endothelial cells (HUVEC) in vitro (example 1). The specification also teaches that overexpression of sphingosine kinase alters adhesion molecule expression in HUVEC, enhances neutrophil adhesion to endothelial cells and promotes tube formation or the endothelial cells arrange into a capillary like network (tubes) in vitro, (example 2). The specification speculates that said tube formation in vitro correlates to angiogenesis and

angiogenesis is a characteristic feature of many chronic inflammatory diseases (specification p 61). While the specification provides teachings pertaining to the effects of overexpression of SK in cells in vitro, the specification fails to provide any relevant teachings or specific guidance or working examples with regard to overexpression of SK in vivo, by introducing a nucleic acid encoding SK, resulting in the modulation of one or more endothelial cell characteristics or in the treatment and/or prophylaxis of a condition characterized by aberrant or otherwise unwanted endothelial cell function in a mammal. The guidance provided by the instant specification fails to correlate the overexpression of SK protein in vitro to overexpression of SK in a cell in vivo resulting in the modulation of an endothelial cell functional characteristic and in treatment and/or prophylaxis of a condition in vivo. Thus, as enablement requires the specification to teach how to make and use the claimed invention, the specification fails to enable the claimed methods for treating and/or prophylaxis of a disease. It would have required undue experimentation to make and use the claimed invention without a reasonable expectation of success.

The claims embrace overexpression of SK in an endothelial cell in vivo by way of introducing a nucleic acid encoding SK resulting in the modulation of endothelial cell characteristics or in the treatment and/or prophylaxis of a condition characterized by aberrant or unwanted endothelial cell functioning, thus falls into the realm of gene therapy. At the time of filing the art taught that gene therapy was unpredictable without undue experimentation. With regard to gene therapy, while progress has been made in recent years for gene transfer in vivo, vector targeting to desired tissues in vivo continuous to be a difficulty as supported by teaching in the art. **Gnewuch et al**, (Cell Mol Life Sci, 59: 959-1023, 2002 (IDS)) while reviewing the status of gene therapy notes there have been several drawbacks to gene therapy including the inaccurate delivery of the gene to the desired cellular localization, the inaccurate transposition of the gene to the required place on the human genome and the lack of activity against

metastasized cancer cells (p 992, 1st bridge 2nd column). Gnewuch et al, assessed gene therapy at that time is not developed to a point of predictable results in all patients (p 992, 2nd column, 2nd paragraph). **Cuvillier** (Anticancer Drugs, 18(2): 105010, 2007 (IDS)) even four years after the filing of the instant application notes sphingosine kinase controls the levels of sphingolipids having opposite effects on cell survival/death, its gene was found to be of oncogenic nature, its mRNA is overexpressed in many solid tumors, its overexpression protects cells from apoptosis and its activity is decreased during anticancer treatments (abstract). Therefore, SK appears to be a potential therapeutic target in cancer (Cuvillier, abstract). However, while progress has been made in recent years for in vivo gene transfer, vector targeting in vivo to desired organs continued to be unpredictable and inefficient. For example, numerous factors complicate the gene delivery art that could not have been overcome by routine experimentation. These include, the fate of DNA vector itself, volume of distribution, rate of clearance in tissue, the in vivo consequences of altered gene expression and protein function, the fraction of vector taken up by the target cell population, the trafficking of the genetic material within cellular organelles, the rate of degradation of the DNA, the level of RNA produced, the amount and stability of the protein produced, and the protein's compartmentalization within the cell, or its secretory fate, once produced (Eck et al , Goodman & Gilman's The Pharmacological basis of Therapeutics, McGraw-Hill, New York, NY. pp 77-101 , previously cited). Cell cultures, which is an n in vitro system is not directly co relatable to the treatment of a mammal which would be in vivo or in situ.

Second the claims embrace the introduction of the nucleic acid encoding SK into an endothelial cell in vivo resulting in modulating endothelial cell functional characteristics or treatment and/or prophylaxis of a condition associated with unwanted endothelial cell functioning. At the time of filing the art is teaching that it is unpredictable if an endothelial cell

would tolerate overexpression of sphinosine kinase in vivo. **Vadas et al,** (Biochimica et Biophysica Acta, 1781: 442-447, 2008) notes in endothelial cells, there is an intolerance of overexpression of SphK1 beyond several folds (p 445, 1st column, 2nd paragraph). The delivery of a vector to target tissue culture cells does not provide guidance cited above for overcoming the obstacles on in vivo delivery because the vector does not have to pass through the complex organization of organs and tissues. Cells cultures do no mimic organs in that there is no three-dimensional structure, blood vessels, connective tissue through which the vector would need to pass in vivo. Any data obtained from cells grown in vitro cannot be extrapolated to the in vivo situation.

The instant specification does not provide any relevant teachings, specific guidance, or working examples for overcoming the limitations of SK gene transfer in vivo resulting in the modulation of endothelia cell in vivo or in the treatment and/or prophylaxis of a disease raised by the state of the art. The art teaches that in vivo models have served as important vehicles to explore a variety of phenotypes associated with metastatic progression and they will continue to do so until the time comes when an in vitro system is developed that faithfully replicates all of the myriad steps and challenges that disseminating tumor cells face (**Khanna et al,** (Cardiogenesis, 26(3): 513-523, 2005) (p 518, 2nd column, last paragraph). Because of the complexity of the metastatic process and the changing microenvironmental cues and interactions that a disseminated cell experiences, the development of such an in vitro assay system is unlikely in the near future (p 518, 2nd column, last paragraph). In vivo models, therefore, must continue to be an important workhorse in metastasis research. Selection of an in vivo model must be tailored to the nature of the question being asked and with full knowledge of the caveats and inadequacies of each model system (p 521, 21st column, 1st paragraph). These models, in conjunction with in vitro modeling and manipulation of tumor cells, have enabled and

will continue to enable investigators to explore the critical questions that remain, including the true nature of metastatic dormancy, the role and identity of the microenvironment cues and the development of agents that can be used to prevent or treat overt metastatic disease (p 521, 21st column, 1st paragraph). The instant specification does not provide any relevant teachings, specific guidance, or working examples for overcoming the limitations of SK gene transfer in vivo resulting in the modulation of endothelia cell in vivo or in the treatment and/or prophylaxis of a disease raised by the state of the art. Given the lack of guidance in the specification and in view of the teachings in the art at the time of filing regarding sphingosine kinase overexpression in vivo, and SK treatment and/or prophylaxis via SK gene therapy, the skilled artisan would require engaging in an undue amount of experimentation without a predictable degree of success to implement in the invention as claimed.

Therefore, in view of the quantity of experimentation necessary to determine the parameters listed above for overexpression of SK by introducing a nucleic acid encoding SK in an endothelial target cell resulting in the modulation of endothelial cell functional characteristics and particularly resulting in the treatment and/or prophylaxis by SK gene therapy, the lack of direction or guidance provided by the specification for overexpression of SK by introducing a nucleic acid encoding SK in an endothelial target cell resulting in the modulation of endothelial cell functional characteristics and particularly resulting in the treatment and/or prophylaxis by SK gene therapy, the absence of working examples that correlate the overexpression of SK by introducing a nucleic acid encoding SK in an endothelial target cell resulting in the modulation of endothelial cell functional characteristics and particularly resulting in the treatment and/or prophylaxis by SK gene therapy, the undeveloped state of the art pertaining to for overexpression of SK by introducing a nucleic acid encoding SK in an endothelial target cell resulting in the modulation of endothelial cell functional characteristics and particularly resulting

in the treatment and/or prophylaxis by SK gene therapy, and the breadth of the claims directed to all diseases associated with aberrant or unwanted endothelial cell functioning, it would have required undue experimentation for one skilled in the art to make and/or use the claimed invention.

Applicants argue that the presently amended claimed methods are directed to the overexpression of sphingosine kinase in endothelial cells and not the overexpression of sphingosine kinase in any cell. The Action alleges that the claims embrace introducing a sphingosine kinase nucleic acid in any type of cell in vivo in order to modulate endothelial cell characteristics or treat and/or provide prophylaxis for a condition characterize by unwanted endothelial cell function. In response these arguments are persuasive that the invention is directed to endothelial cells, however, as discussed below applicants have not provided guidance for a method for modulating mammalian endothelial cell functional characteristics by way of the claimed methods in vivo.

Applicants argue no working examples are required to satisfy the enablement requirement.

Applicants argue the specification need not contain an example if the invention is otherwise disclosed in such manner that one skilled in the art will be able to practice it without an undue amount of experimentation. Applicants argue the as-filed specification provides many working examples of the presently claimed invention. Applicants respectfully submit that the as-filed specification provides ample guidance for the expression of a sphingosine kinase in endothelial cells. For example, the as-filed specification teaches that overexpression of sphingosine kinase using an adenoviral vector in vitro leads to increased endothelial cell survival in adenoviral treated cells compared to control cells (see Example 1 of the as-filed specification). Furthermore, endothelial cells treated in vitro with adenoviral sphingosine kinase display lower

levels of caspase-3 activity compared to control cells (see Example 1 of the as-filed specification). Example 2 of the as-filed specification shows that endothelial cells treated in vitro with adenoviral sphingosine kinase display increased vascular cell adhesion molecule expression, increased neutrophil adhesion, and vascular tube formation. Applicants argue that these in vitro examples correlate with the presently claimed methods. Applicants reiterate that the use of in vitro experiments to establish in vivo events is, in principle, a valid methodology.

These arguments are not persuasive because an artisan cannot correlate the in vitro guidance provided in the specification to the in vivo. The art teaches that sphingosine kinase expression in an endothelial cell in vitro cannot be correlated to its expression in vivo, as **Vadas et al**, notes for example, in endothelial cells, there is an intolerance of overexpression of SphK1 beyond several folds (p 445, 1st column, 2nd paragraph). As such it is unpredictable what levels of sphingosine kinase transgene expression are required in vivo in order to modulate one or more endothelial cell functional characteristics in vivo. This is underscored by the teachings of **Cuvillier** who notes sphingosine kinase controls the levels of sphingolipids having opposite effects on cell survival/death, its gene was found to be of oncogenic nature, its mRNA is overexpressed in many solid tumors, its overexpression protects cells from apoptosis and its activity is decreased during anticancer treatments (abstract). However, while progress has been made in recent years for in vivo gene transfer, vector targeting in vivo to desire organs continued to be unpredictable and inefficient. For example, numerous factors complicate the gene delivery art that could not have been overcome by routine experimentation. These include, the fate of DNA vector itself, volume of distribution, rate of clearance in tissue, the in vivo consequences of altered gene expression and protein function, the fraction of vector taken up by the target cell population, the trafficking of the genetic material within cellular organelles, the rate of degradation of the DNA, the level of RNA produced, the amount and stability of the

protein produced, and the protein's compartmentalization within the cell, or its secretory fate, once produced (Eck et al , Goodman & Gilman's The Pharmacological basis of Therapeutics, McGraw-Hill, New York, NY. pp 77-101). Thus, cell cultures, which is an n in vitro system is not directly co relatable to the treatment of a mammal. Therefore, one skilled in the art would accept the sphingosine kinase in vitro model as reasonably correlating to sphingosine kinase activity model for in vivo applications. Based upon the relevant evidence as a whole, there is no reasonable correlation between the disclosed in vitro sphingosine activity and an in vivo activity. Therefore a correlation is necessary where the disclosure of sphingosine kinase activity in vivo activity is reasonable for sphingosine kinase gene therapy.

Applicants argue that there are no three dimensional structures, blood vessels, or connective tissues through which the nucleic acid would be required to cross through in vivo to effect therapy. Adenovirus is routinely administered via the vasculature which contacts all endothelial cells. In fact, some types of endothelial cells are part of the vasculature (e.g., vascular endothelial cells). The art of gene therapy at the time the application was filed had successfully solved the problem of delivering adenoviral constructs to endothelial cells. Applicants respectfully submit herewith, examples of adenoviral administration to endothelial cells, wherein in vitro cell culture models correlate to the in vivo models. See, for example, Lemarchand et al. Circulation Research 1993, 72, 1132-1138; Schulick et al. Circulation Research 1995, 77, 475-485; Budenz et al. Investigative Ophthalmology and Visual Research, 1995, 36, 11,2211-2215; Zoldhelyi et al. Circulation 1996, 93, 10-17; White et al. Hypertension 2001, 37, 449-455; Champion et al. Circulation Research, 1999, 84, 1422-1432; and Claudio et al. Circulation Research 1999, 85, 1032-1039; a copy of each reference is attached for your convenience.

These arguments are not persuasive because the instant invention does not require adenovirus but requires an isolated nucleic acid which is required to reach an endothelial cell in vivo by passing through three dimensional structures, blood vessels, or connective tissues in vivo to effect therapy. In addition it requires the isolated nucleic acid to be taken up by the target endothelial cell, the trafficking of the genetic material within cellular organelles, the rate of degradation of the DNA, the level of RNA produced, the amount and stability of the protein produced, at sufficient effective amounts for modulating endothelial cell functional characteristics or for the treatment and/or prophylaxis of a condition. Lemarchand et al. in vivo gene transfer and expression in normal uninjured blood vessels using replication-deficient recombinant adenovirus with LacZ or human anti-trypsin fails to provide guidance for nucleic acid sphingosine kinase gene therapy. This is because each transgene requires different levels of transgene expression sufficient to have an effect in vivo. Moreover, Lemarchand et al. teaches detection of the transgene expression at the target cell and not a therapeutic effect as required in the instant invention.

Applicants argue Duan et al., 2007, previously made of record, provides post-filing examples of the reduction to practice of adenoviral sphingosine kinase administration in mammals. In addition, the reference correlates in vitro observations to support their studies. In fact, Duan et al. expressed sphingosine adenovirus in isolated rat cardiac myocytes, isolated rat hearts, and in vivo rat hearts. However, the Examiner objected to Duan because the effects allegedly involved cardiac muscle cells and not endothelial cells. Applicants respectfully submit that Duan et al., applies to the principle that in vitro models of adenoviral shpingosine kinase reasonably correlate with in vivo models. To further bolster this point, Applicants submit a post-filing example, Lee et al., Coronary Artery Disease, 2005, 16, 451-456, (copy attached) which describes the use of adenoviral sphingosine kinase to promote arteriogenesis in a rabbit

hindlimb ischemia model. Lee et al. provide a correlation of an in vitro endothelial cell model with an in vivo endothelial cell model using sphingosine kinase, as disclosed in the as-filed specification.

The Action further contends that in vitro studies of a protein's function at the cellular level is problematic due to interactions with other molecules and precludes the studies of physiological (e.g., metabolic pathways) and phenotypic functions in a mammal (e.g., role of the protein in the whole mammal). Applicants fail to understand the Examiner's rationale. Such rationale would preclude the correlation of any in vitro assay to an in vivo assay, unless the assay environments were indistinguishable. Applicants respectfully submit that this rationale is not only impractical, but irrelevant.

These arguments are not persuasive because as discussed above the targeting an endothelial cell in vivo with an adenovirus is distinct from targeting an endothelial cell with an isolated nucleic acid. For example, an adenovirus enters a target endothelial cell via the adenovirus receptors while a nucleic acid is required to cross the plasma membrane to enter the target endothelial cell. This differential cellular uptake requires different levels of the nucleic acid administration as compared to adenovirus and moreover, results in differential effective amounts of the sphingosine kinase product for an effect in vivo. For a claimed genus of sphingosine kinases, one skilled in the art would have to perform an undue experimentation to decide the levels of nucleic acid administration, and the effective amounts of transgene expression resulting in modulating endothelial functional characteristics and in particular resulting in treating and/or prophylaxis of a condition as instantly claimed.

Double Patenting

The nonstatutory double patenting rejection is based on a judicially created doctrine grounded in public policy (a policy reflected in the statute) so as to prevent the unjustified or

improper timewise extension of the "right to exclude" granted by a patent and to prevent possible harassment by multiple assignees. A nonstatutory obviousness-type double patenting rejection is appropriate where the conflicting claims are not identical, but at least one examined application claim is not patentably distinct from the reference claim(s) because the examined application claim is either anticipated by, or would have been obvious over, the reference claim(s). See, e.g., *In re Berg*, 140 F.3d 1428, 46 USPQ2d 1226 (Fed. Cir. 1998); *In re Goodman*, 11 F.3d 1046, 29 USPQ2d 2010 (Fed. Cir. 1993); *In re Longi*, 759 F.2d 887, 225 USPQ 645 (Fed. Cir. 1985); *In re Van Omum*, 686 F.2d 937, 214 USPQ 761 (CCPA 1982); *In re Vogel*, 422 F.2d 438, 164 USPQ 619 (CCPA 1970); and *In re Thorington*, 418 F.2d 528, 163 USPQ 644 (CCPA 1969).

A timely filed terminal disclaimer in compliance with 37 CFR 1.321(c) or 1.321(d) may be used to overcome an actual or provisional rejection based on a nonstatutory double patenting ground provided the conflicting application or patent either is shown to be commonly owned with this application, or claims an invention made as a result of activities undertaken within the scope of a joint research agreement.

Effective January 1, 1994, a registered attorney or agent of record may sign a terminal disclaimer. A terminal disclaimer signed by the assignee must fully comply with 37 CFR 3.73(b).

Claims 1-3, 5, **stand** rejected on the ground of nonstatutory obviousness-type double patenting as being unpatentable over claims 1-6, 15-20 of U.S. Patent No. 10275,686. Applicants argue that until time the claims are in condition for allowance, Applicants will submit a terminal disclaimer.

Claims 1-2, 5-7, 15 **stand** rejected on the ground of nonstatutory obviousness-type double patenting as being unpatentable over claims 1-15, 17, 23, of U.S. Patent No. 09/977,217. Applicants argue that until time the claims are in condition for allowance, Applicants will submit a terminal disclaimer.

Conclusion

No claim is allowed.

THIS ACTION IS MADE FINAL. Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO

MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will the statutory period for reply expire later than SIX MONTHS from the mailing date of this final action.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Magdalene K. Sgagias whose telephone number is (571) 272-3305. The examiner can normally be reached on Monday through Friday from 9:00 am to 5:00 pm. If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Peter Paras, Jr., can be reached on (571) 272-4517. The fax phone number for the organization where this application or proceeding is assigned is (703) 872-9306.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll free).

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Art Unit 1632

/Anne-Marie Falk/
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